# Considerations for Development of Entrained Oil Thresholds for Oil Spill Risk Assessments

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## Summary

Oil spill modelling using full oil fate models such as RPS/TetraTech's SIMAP model provides output for two types of water column concentration data: (1) dissolved oil compounds and (2) total hydrocarbons and related compounds in petroleum oils (abbreviated as THC) in entrained oil droplets. The purpose of this technical note is to identify appropriate thresholds for use in oil spill risk assessments. The focus is on developing a THC threshold appropriate for modelled entrained oil droplets.

As described in detail herein, toxicity is related to the bioavailability of hydrocarbons and related compounds in petroleum oils (described as "hydrocarbons" herein for brevity). Soluble and semi-soluble components of oil dissolve and therefore become bioavailable (i.e., able to be taken up by aquatic organisms). In relatively fresh oil, some of the hydrocarbons in entrained oil droplets are soluble/semi-soluble compounds that may later dissolve and become bioavailable. However, as oil weathers, these potentially toxic components diminish to the point where the THC in entrained droplets is effectively non-toxic. Therefore, the focus of a risk assessment is typically on the dissolved hydrocarbon exposure.

Measurement of the concentration of oil in water, whether in the field or in laboratory studies, is complicated and not all constituents can be identified and measured individually. Mass recoveries (using the presently recommended methods) for gasoline, kerosene, and gas oil are typically >95%, whereas the mass recoveries for heavier substances in crude oils and heavier refined products are often <30%. As only some of the compounds in oil are measurable individually, and because of the desire to be inclusive of effects from hydrocarbons in oil droplets as well as the dissolved phase, aquatic toxicologists have attempted to use THC as a metric for evaluating toxicity of oil exposures. However, given the variable composition of oil as it weathers, development of effects levels or thresholds based on total hydrocarbons (THC) is problematic. The review series (Loughery et al. 2023; Stubblefield et al. 2023; Alloy et al. 2023; Parkerton et al. 2023a,b; Dettman et al. 2023; Bejarano et al. 2023; Lee et al. 2023; French-McCay et al. 2023) discusses these complications and provides guidance for aquatic toxicity studies of petroleum oils and their application to risk assessments and modelling-based analyses.

For the compounds dissolved from oil to which aquatic biota are primarily exposed (i.e., polycyclic aromatic compounds, PACs), thresholds based on acute lethality (LC50s, i.e., lethal concentrations for 50% of test organisms ) generally range from about 10 ppb (µg/L) for sensitive early life history stages of aquatic species to 300 ppb, or more for less sensitive species

and older life stages, if exposed for more than 2 days. For sub-lethal effects, a threshold of 1 ppb is conservatively protective of aquatic biota where exposure durations are sufficiently long (i.e., typically >10 days) for such effects to occur. For oil spills in open water, the acute threshold of 10 ppb would be appropriate for consideration of sublethal effects, given short-term exposures that occur in the open ocean environment.

If THC in oil droplets entrained in the water column is to be evaluated as a risk or to define an area for monitoring or the Environment that May Be Affected (EMBA, as defined by NOPSEMA, 2025), weathering of entrained droplets should be considered, along with duration of exposure. For freshly spilled oil still containing the soluble and semi-soluble compounds that could later dissolve, 1 ppm (1 mg/L; 1,000 ppb) would be a sufficiently conservative threshold for oil droplets of all oil types and all weathering states, with 3 ppm (3 mg/L; 3,000 ppb) an appropriate threshold for crude oils. For light distillates and condensates, 1 mg/L (1 ppm; 1,000 ppb) is an appropriate threshold. This threshold is based on bioassay studies measuring total measurable hydrocarbons in both droplet and dissolved phases involving 2 or more days of exposure to fresh oil dispersions where the dissolved phase caused most of the effects. Effects levels for weathered oil and for shorter duration exposures would be considerably higher (i.e., by an order of magnitude).

## Introduction

An objective of oil spill risk assessments is to evaluate whether aquatic habitats and organisms could be exposed to oil or its constituents at concentrations above levels of concern at some time after a spill. Typically, oil transport and fate modelling (e.g., using RPS/TetraTech's Spill Impact Model Application Package [SIMAP], French-McCay 2003, 2004, French-McCay et al. 2021a)) is used to estimate over space and time: 1) the spread and concentrations of oil on the water surface, 2) concentrations of oil (i.e., entrained whole oil droplets) in the water column, 3) dissolved concentrations in the water column, and 4) standing of oil on shorelines. Floating and shoreline oil thresholds have been based on observed effects after spills. Water column concentrations have been compared to thresholds based on laboratory toxicity studies performed under controlled conditions designed to provide continuous exposures (e.g., typically without dilution and water column mixing) over relatively long durations (typically 4 days) at sufficiently elevated concentrations to induce a measurable response. These relatively long exposure times have been adopted to allow time for organisms to absorb oil components.

In Australia, recent practice, based on government (National Offshore Petroleum Safety and Environmental Management Authority [NOPSEMA]) guidance (NOPSEMA, 2019, 2025), has been to identify all areas exceeding conservative threshold concentrations at any instant in time after a spill as the Environment that May Be Affected (EMBA). An EMBA is not defined as an area where adverse impacts would be expected throughout the area, but rather where more analysis is to be performed to determine if impacts could occur or have occurred (i.e., if there is a spill) to some resources and/or their uses. The proponent is required to develop a scientific monitoring program for the EMBA's extent, as well as consult with stakeholders regarding potential impacts or concerns.

This review document is an update to the technical note I prepared (French-McCay, 2025), which reviewed the NOPSEMA (2019) guidance. New guidance was published by NOPSEMA (2025) in December of 2025. In addition to updating this document based on the 2025 NOPSEMA guidance, additional literature related to the potential effects of entrained oil droplets was reviewed and summarized herein.

Based on NOPSEMA (2025) guidance, in determining the EMBA and the operational and scientific monitoring planning area, consideration is to be given to four oil pathways: floating oil, entrained oil droplets, dissolved aromatic hydrocarbons, and stranded oil (on shorelines). The "low" (i.e., conservative) thresholds recommended by NOPSEMA (2025) for the operational and scientific monitoring planning area are:

- Floating oil as THC: 1 g/m$^2$ = ~1,000 liters/km$^2$
- Shoreline oil as THC: 10 g/m$^2$ = ~10 mL/m$^2$
- Dissolved oil as polycyclic aromatic hydrocarbons (PAH): 10 µg/L (~10 ppb)
- Entrained oil as THC:
    - 10 µg/L (~10 ppb) for defining the operational and scientific monitoring planning area (within which further analysis can be done to inform monitoring priorities)
    - 100 µg/L (~100 ppb) for defining the EMBA

The floating and shoreline oil thresholds are consistent with the literature reviews and analyses described in French McCay (2016) and French McCay et al. (2018). Based on the Bonn Agreement Oil Appearance Code (BAOAC, Bonn Agreement, 2009, 2011), the floating oil threshold is the typical thickness of rainbow sheen. Potential for effects on socioeconomic resources may occur (e.g., fishing may be prohibited) if oil is visible on the water surface, i.e., sheens. Effects on socioeconomic resource uses may occur (e.g., recreation, tourism) above the shoreline threshold, such that this level of contamination may trigger the need for shoreline clean-up on amenity beaches. Thus, these are reasonable thresholds for establishing an EMBA.

While the dissolved threshold of 10 ppb is consistent with analyses in French McCay (2016) and French McCay et al. (2018), those publications did not advocate for 10 ppb as an entrained oil threshold. Entrained oil is whole oil mixed into the water column, whereas the compounds in oil that are soluble or semi-soluble and can dissolve represent a small fraction of whole oil. Thus, the entrained oil threshold should be much greater than that for dissolved. French McCay (2016) had argued that since the dissolved concentrations in the water column that cause toxicity are primarily polycyclic aromatic hydrocarbons (referred to as PAHs, but more generally and accurately termed polycyclic aromatic compound, PACs, which include PAHs and related cyclic compounds) that typically represent about 1% of the oil when fresh (Meador and Nahrgang, 2019), and these components are lost from the oil as it weathers, an entrained oil threshold for fresh oil should be at least 100 times that for dissolved concentrations. More generally, crude oils contain 1-3%, and condensates contain up to 7% of these measurable PAHs. While the entrained oil threshold was said to be based on potential for exceedance of water quality guidelines (ANZECC/ARMCANZ, 2000; NOPSEMA, 2025), it is unclear what the precise basis of the 10-ppb value would have been.

In defining the EMBA and operational and scientific monitoring planning area, Environment Plans are to consider any probability (i.e., even one trajectory out of hundreds of potential pathways) of exceeding the NOPSEMA-recommended threshold for any of the four oil pathways at any instant in time. Consequently, recent EMBAs have encompassed large areas of the oceans surrounding Australia, owing to use of these conservative thresholds developed without consideration of the dynamic nature of an oil spill (i.e., dilution and dispersion in 3-dimensions and time), duration of exposure, the degree of weathering of the entrained oil, the relative toxicity of different contaminants in oil, and the portion of the water column potentially exposed. Most commonly the extent of the EMBA has been controlled by the distance that the threshold for entrained oil is exceeded instantaneously, with this distance being much larger than the distance that the threshold for dissolved is exceeded instantaneously. This reveals that most EMBA bounds have extended beyond where entrained oil could be a source of toxic soluble and semi-soluble components. An unintended consequence of using an overly conservative threshold is that resources for assessing impacts (e.g., water sampling, monitoring) would be spread across a large area, diminishing the effectiveness of these efforts.

Thus, of particular concern are the 10 ppb threshold and 100 ppb threshold for entrained oil recommended by NOPSEMA (2025) to define the area for scientific monitoring and the EMBA, respectively. The use of these thresholds does not consider the bioavailability of constituents in the oil that drive aquatic toxicity, the changing composition of oil as it weathers, or the likelihood and duration of exposure.  Oil fate and exposure modelling addresses the complexities of oil composition, weathering, partitioning in the environment, and organisms' behaviors. In view of these points, more appropriate entrained oil thresholds and considerations could be used for risk evaluations and establishing an EMBA, as described below.

## Bioavailability

### Dissolved Compounds in Oil are Bioavailable

Dissolved compounds are taken up into organisms directly through external surfaces and gills, as well as through the digestive tract. Thus, soluble and semi-soluble hydrocarbons and related compounds in oil are bioavailable, whereas insoluble compounds in oil are not bioavailable to aquatic organisms. Laboratory studies have shown that the oil constituents dissolved in the water (or media) exert the most effects on aquatic biota (Carls et al. 2008; Nordtug et al. 2011b; Olsvik et al. 2011; Redman 2017a; Gobas et al. 2018).

The fraction of oil with the most toxic effects to water-column and benthic organisms are lower-molecular-weight compounds (i.e., PACs, often referred to as PAHs), which are volatile, and soluble or semi-soluble in water. As a result, they persist in the environment long enough for significant exposure to occur (Anderson et al., 1974, 1987; Neff and Anderson, 1981; Malins and Hodgins, 1981; McAuliffe, 1987; NRC 2003, 2005; Nordborg et al. 2018).  The monoaromatic hydrocarbons (MAHs), including BTEX (benzene, toluene, ethylbenzene, and xylenes) and substituted benzenes, and the soluble alkanes also contribute to toxicity, but these compounds are highly volatile, so exposures of aquatic biota are minimal or negligible except when light oils are discharged at depth where volatilization does not occur (French-McCay 2002).

Within the soluble and semi-soluble hydrocarbons, toxicity is inversely related to solubility, typically quantified by the octanol-water partition coefficient ($K_{ow}$), a measure of hydrophobicity (Nirmalakhandan and Speece 1988; Hodson et al. 1988; Blum and Speece 1990; McCarty 1986; McCarty et al. 1992a, b; Mackay et al. 1992; McCarty and Mackay 1993; Verhaar et al. 1992, 1999; Swartz et al. 1995; French-McCay 2002; McGrath et al 2009). The range of LC50s varies from ~10 µg/L (ppb) for 3-ring PAHs (which are semi-soluble) to ~10-100 mg/L (ppm) for the highly soluble BTEX compounds (French-McCay 2002). Thus, the toxicity of an oil hydrocarbon mixture is strongly related to the chemical composition, which varies as the oil weathers since the soluble and semi-soluble hydrocarbons are all volatile to varying degrees.

### Oil Droplets

While entrained oil droplets could potentially affect organisms by mechanical means (i.e., fouling, interference with feeding), research has not definitively demonstrated direct effects of droplets as separable from the effects of components dissolved from the droplet oil (Carls et al. 2008; Hansen et al., 2018, 2019a,b; Parkerton et al. 2023b). Contact of droplets with organisms' surfaces (external or by ingestion) could facilitate direct exposure to soluble and semi-soluble components if they become bioavailable (i.e., dissolve and enter the organism though external or internal membranes; Letinski et al. 2014; Redman et al. 2017a; Hansen et al. 2018, 2025). Limited contribution of oil droplets to toxicity has been observed for fish larvae, even using continuous exposure to freshly formed oil droplets (i.e., using methods described in Nordtug et al. 2011a) over 3 to more than 10 days (Nordtug et al., 2011b; Olsvik et al., 2011, Hansen et al. 2012, 2015, 2016, 2019a, 2025). Sørhus et al. (2015, 2016, 2017, 2021a,b, 2023a,b), Sørensen et al. (2017, 2019), and Hansen et al. (2018) have shown that uptake of PACs can occur if oil micro-droplets adhere to fish eggs with sticky surfaces (i.e., the chorion). However, as noted by Hansen et al. (2019a), the potential contribution of oil droplets to toxicity remains to be quantified, but should be considered. While quantification in experimental systems and in the field is challenging, oil spill models such as SIMAP (French-McCay 2003, 2004, French-McCay et al. 2021a) and OSCAR (Reed et al. 2004) calculate dissolution from oil droplets into the dissolved phase, so the contribution of oil droplets via this exposure route is included when evaluating exposure to dissolved concentrations.

Further discussion of toxicity studies involving exposures to oil droplet dispersions is below in the Section: Experiments Exposing Organisms to Oil Droplet Dispersions. A first attempt at modelling the adherence of oil micro-droplets to (assumed adhesive) fish eggs by Frøysa et al. (2025) is also discussed in that section.

## Oil Composition Affects Toxicity

Oil is a complex mixture of thousands of hydrocarbons and related compounds of varying physical, chemical, and toxicological characteristics, and therefore, varying environmental fates and effects on organisms. As discussed above, due to differing solubility, oil components vary by orders of magnitude in their potential to cause adverse effects, due to varying bioavailability. A fraction of oil does not readily biodegrade, because the compounds involved are not bioavailable.

Oils differ in composition and their fractions of potentially bioavailable components, and so not all oils are equally toxic.

Thus, the use of a simple sum of components to quantify toxicity is problematic, as the composition of dissolved components to which organisms are exposed in the field will differ from that in the laboratory study used to develop the toxicity threshold. Toxic effects levels vary by what is included in the summation and the relative concentrations in the mixture.

The approach used to quantify the additive effects of a mixture of hydrocarbons and related compounds is to use a Toxic Unit (TU) model (Swartz et al. 1995; DiToro et al. 2000; French-McCay 2002; Redman et al. 2017b; NASEM 2020; Parkerton et al. 2023a; see review by French-McCay et al. 2023). A TU for evaluating mortality is defined as the exposure concentration divided by the lethal concentration affecting 50% of exposed organisms (LC50). Thus, at TU=1, mortality is expected. For a mixture, the toxic units are additive. When for a mixture of chemicals at specific concentrations $\Sigma$ TU = 1, the mixture is predicted to be lethal to 50% of exposed organisms. Use of the TU approach for evaluating effects addresses the varying composition of oil-derived mixtures, including changes in composition due to weathering. Examples of the use of the TU model for interpreting oil exposures are in Negri et al. (2021), Philibert et al. (2019, 2021), and Parkerton et al. (2023b).

## Oil Weathering

As oil weathers, its composition changes due to volatilization and dissolution losses, and biodegradation (Payne et al. 1984, 1991; Stout et al. 2016; Boehm et al. 2016; Payne and Driskell 2018; Wade et al. 2018). When oil is floating, the volatile components evaporate rapidly, and the oil becomes more viscous as a result. Some of the semi-soluble hydrocarbons, which are less volatile, dissolve from the floating oil into the water column. Floating oil may be entrained into the water column by breaking waves, or oil may be released under water. Soluble and semi-soluble hydrocarbons dissolve from subsurface oil droplets, weathering the oil and making them more available to microorganisms. The uptake of hydrocarbons by microorganisms, referred to as biodegradation, reduces water column concentrations, and therefore toxic effects. The volatilization rates of hydrocarbons from surface slicks are faster than the dissolution rates. Thus, dissolution from oil droplets in the water column is the main source of concentrations dissolved in the water (French-McCay 2002).

Therefore, oil weathering leads to a preferential loss of the toxic components from the whole oil. The more bioavailable soluble and semi-soluble components evaporate and dissolve (and potentially cause acute toxicity in water column), leaving residual oil with lower potential for causing adverse effects. Entrained oil droplets weather rapidly because of their high surface area relative to their volume, owing to their small size. Microbes attach to their surfaces and readily take up soluble and semi-soluble components. Driskell and Payne (2018) provide evidence of these weathering processes being accelerated for smaller dispersant-treated oil droplets in their analyses of water samples collected during the Deepwater Horizon oil spill (DWHOS) in the Gulf of Mexico.

Forth et al. (2017) measured total PAH concentration for 50 bioavailable PAHs and related heterocyclic compounds (TPAH50, components of the oil causing the most toxicity) in fresh MC252 (DWHOS) light crude oil (API 36) and weathered samples. Fresh MC252, artificially weathered, slick A, and slick B oils contained 1.06%, 1.38%, 0.31% and 0.33 TPAH50, respectively. The artificially weathered, slick A, and slick B oils were 27%, 68% and 85% depleted in TPAH50, based on ratios to C30-hopane (which does not dissolve or degrade). The artificially weathered oil (produced by heating fresh oil in the lab) had a mass loss of approximately 33% to 38% relative to the fresh oil, removing the BTEX but concentrating the TPAH50 slightly. Slick A originated from oil collected by various skimmer vessels near the MC252 well. Slick B was collected by a skimmer vessel on 19 July 2010, 4 days after the oil release was stopped. Thus, more depletion of TPAH50 from the oil occurred as oil remained at sea and moved away from the source.

TPAH50 depletions averaged 69 ± 23% for DWHOS floating oils analyzed by Stout et al. (2016). The samples were collected between 1.5 and 69 km from the MC252 source, but mostly less than 25 km from the wellhead. Samples taken more than 10 km from the well were >70% depleted in TPAH50. Floating oil samples collected less than 10 km from the wellhead were 12 to 91% depleted in TPAH50, averaging 54 ± 20%. The variability was due to newly surfacing oil mixing in with more weathered older oil with much lower TPAH50 content due to rapid evaporation.

Figure 1 provides an example of the effects of oil weathering for floating oil from the DWHOS, as modelled by French-McCay et al. (2021a,b). While the oil and gas release from 1500 m continued until 15 July 2010, freshly released oil was mixed with weathered oil, such that after the first few days of the spill, the oil on average contained <1% of soluble and semi-soluble components (i.e., BTEX, substituted (C3- and C4-) benzenes, PAHs and heterocycles). The PAHs and heterocycles (i.e., TPAH50) remain in the water much longer than the highly volatile and soluble MAHs; hence contribute most to aquatic toxicity.

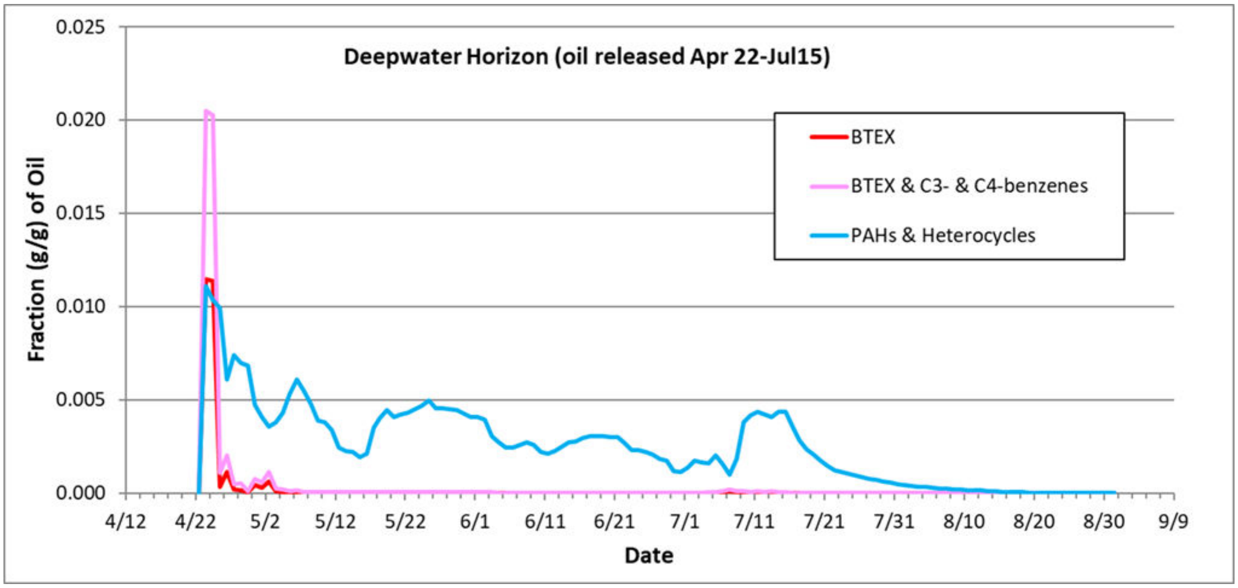

Figure 1. Average fraction of oil represented by soluble and semi-soluble aromatic components over time for the DWHOS, as modelled by French-McCay et al. (2021a,b).

Boehm et al. (2016), Payne and Driskell (2017, 2018), Driskell and Payne (2018), and Wade et al. (2018) evaluated water sample chemistry results from the DWHOS field monitoring collections overseen by the US National Oceanic and Atmospheric Administration (NOAA). The results showed considerable weathering with distance from the well, i.e., with time in the water column. The MAHs were shown to be rapidly depleted from the entrained oil droplets before the oil reached the surface. Semi-soluble PAHs were depleted both as the oil rose through the water column (Boehm et al. 2016; Payne and Driskell 2017, 2018; Driskell and Payne 2018; Wade et al. 2018) and as it moved away from the source at the surface (Stout et al. 2016). Further, the compositional PAH profiles in the subsurface water samples allowed for the quantification of the fraction of PAHs in droplets versus dissolved phases (see methods in Payne and Driskell 2017, 2018). The droplets were highly weathered and contained much lower concentrations of TPAH50 than the fresh MC252 oil (Payne and Driskell 2017, 2018; Driskell and Payne 2018).

Highly weathered petroleum residuals have not been shown to have adverse effects on aquatic organisms, i.e., they are not acutely toxic in the water column. Therefore, effects levels for the bioavailable soluble and semi-soluble components should not be applied to whole oil droplets entrained in the water column, particularly for weathered entrained oil droplets.

## Consideration of Duration of Exposure

### Time for Uptake of Hydrocarbons

An additional consideration is that LC50s and other toxicity endpoints vary considerably with exposure duration over the range of several hours to several days (Sprague, 1969; Abel, 1980; Mancini, 1983; Bailey et al., 1985; McAuliffe 1987; McCarty et al. 1992a, b). Effects thresholds are an order of magnitude higher for a few hours of exposure than they are for several days of exposure (McCarty et al. 1992a, b; French-McCay 2002; Lee et al. 2002; Bejarano et al. 2014; Mackay et al. 2017) due to the hydrocarbon accumulation over time up to a critical tissue concentration that causes mortality. Exponentially higher concentrations are required to achieve equivalent effects over shorter durations (e.g., Figures 2 and 3). The accumulation is slower for more hydrophobic (i.e., less soluble) compounds (Veith et al., 1983; Abernethy et al. 1986, 1988; Verhaar et al., 1992, 1999; DiToro et al. 2000; DiToro and McGrath 2000; see reviews in McCarty et al. 1992a, b; Verhaar et al., 1992; French-McCay 2002; French-McCay et al. 2023). Thus, the LC50s for PAHs are more variable with duration of exposure than are the LC50s for BTEX.

Most bioassays are performed under 48 to 96 hours of exposure to allow concentrations to accumulate in the tissues of the exposed organisms, where they may have adverse effects. Swartz et al. (1995) argued that bioassays for PAHs should be to at least 10 days of exposure in order to allow sufficient time for the PAHs to be taken up into the organisms and reach steady state between uptake and depuration. The focus on Swartz et al.’s (1995) study was PAH contamination in sediments. For long, essentially infinite exposure durations, the measured lethal endpoints are termed incipient LC50s (or other percentage mortality levels; McCarty et al. 1992a, b; Verhaar et al., 1992; French-McCay 2002). The >48-hour and incipient LC50s are conservatively low for shorter-duration exposures (French-McCay 2002). Animals can be unaffected by short-duration exposures to concentrations that could affect them if exposed for days or weeks. Organisms can recover from sublethal exposures, by elimination of toxins from tissues. Taking these considerations into account, application of thresholds developed from exposure periods of days or weeks are highly conservative when applied to an instantaneous exposure.

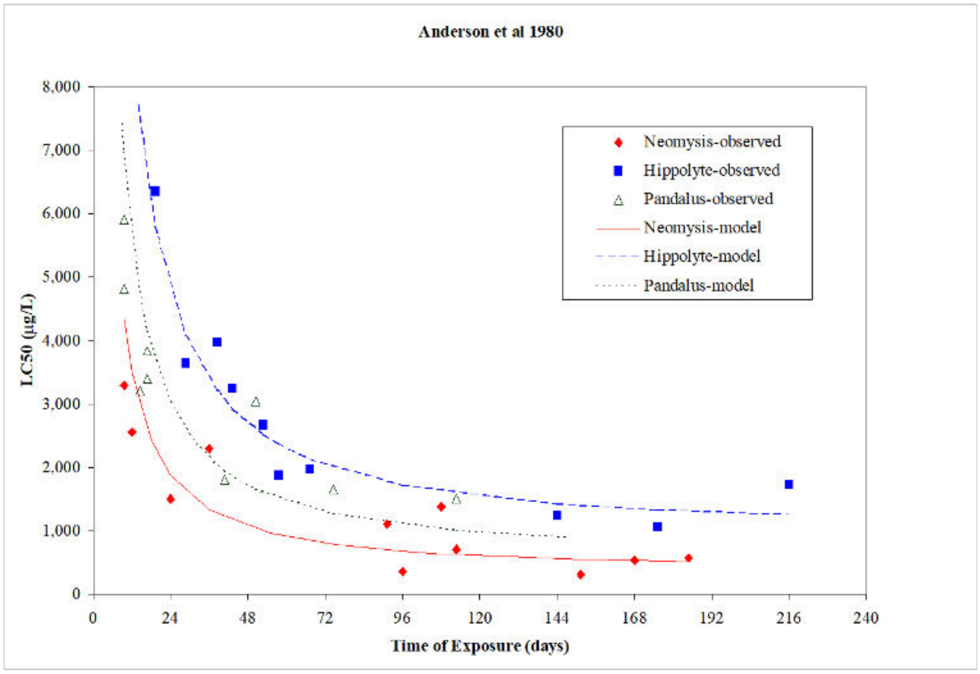

Figure 2. Lethal concentrations (LC50) versus duration of exposure in experiments reported by Anderson et al. (1980).

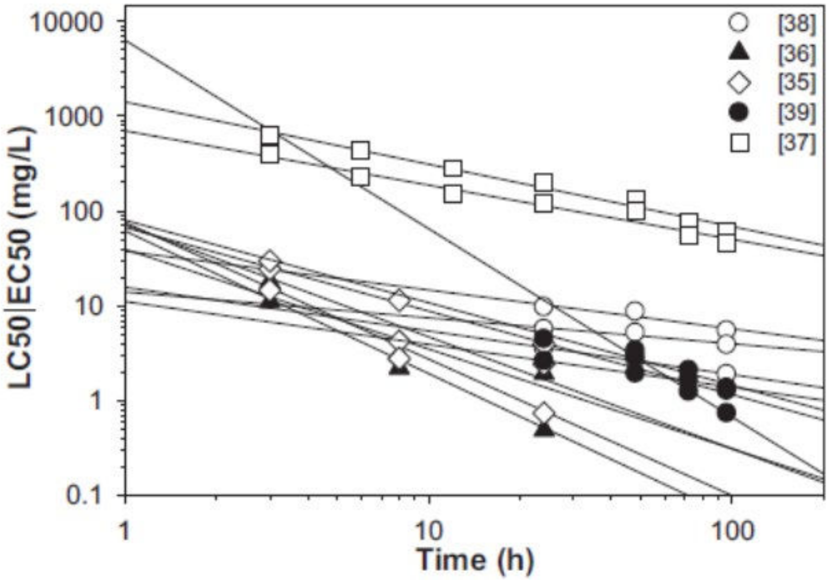

Figure 3. Lethal and effects concentrations affecting 50% of test organisms (LC50, EC50) versus duration of exposure in experiments reviewed by Bejarano et al. (2014; see source for original citations).

## Oil Dynamics

For most oil spills, exposures of water column biota to concentrations above potential thresholds of concern are typically on time scales of minutes to hours, even for spills lasting weeks or months because of the varying movements of the oil in the water, dilution and losses to biodegradation and volatilization. Furthermore, the concentrations vary in time over the short exposure periods (McAuliffe et al. 1980, 1981; McAuliffe 1987; Lunel 1994; French McCay 2002, 2004; Bejarano et al. 2014; French-McCay et al. 2023).

Marine and coastal waters are not static systems, as the oil rapidly spreads and moves through space over time. Oil is concentrated floating on the surface, along shorelines, and as it settles to sediments, while it is dispersed (diluted) in three-dimensional space in open water. When floating oil is entrained, a portion refloats immediately, more so for larger droplets originating from more viscous oils. Entrained oil in smaller droplets than those that refloat, rapidly disperses into the water column away from the surface. Currents and dispersion in the water column, combined with differential buoyancy of oil droplets of various sizes, rapidly dilute concentrations of entrained droplets. Therefore, oil exposures in open water are typically of short duration.

While it is recognized that in offshore open water, oil exposures are ephemeral, toxicologist have expressed concerns that exposures would be much longer where oil may accumulate. Embayments in coastal areas can act as low-energy, low-mixing environments where hydrocarbons may accumulate and persist (Hook 2020). However, to accumulate the entrained oil would need to surface, come ashore or settle to sediments in these areas. While oil is entrained in the water, the concentrations do not physically concentrate, rather they dilute by turbulent diffusion. The SIMAP model evaluates entrained oil surfacing and accumulation of oil at the surface and in sediments, such as in nearshore areas (which occurs along shorelines). Exposure concentrations for floating and shoreline oil address these potential exposures. Use of floating and shoreline oil thresholds identify areas where there would be potential for oil accumulation and potential impacts on sensitive species and their life stages. Furthermore, floating oil and entrained oil do not move in the same manner, as the floating oil is transported by wind effects on the water surface as well as by surface currents, whereas entrained oil moves with subsurface currents. Thus, the entrained oil “footprint” is not indicative of where oil might accumulate.

Figure 4 provides an example of the changing composition of dissolved hydrocarbons and the short duration of exposure to high concentrations in a tank containing floating crude oil held in place by the containment (Payne et al. 1984). As the oil weathered, concentrations below the slick decreased and became dominated by PAHs. If the floating oil were in the ocean, the dissolved components would be spread by the movement of the oil over the surface and by current shear, such that concentrations would be much lower, more ephemeral, and patchy. Bejarano et al. (2014) reviewed measurement data from field trials finding oil concentrations decreased rapidly (in minutes) after oil release (Figure 5).

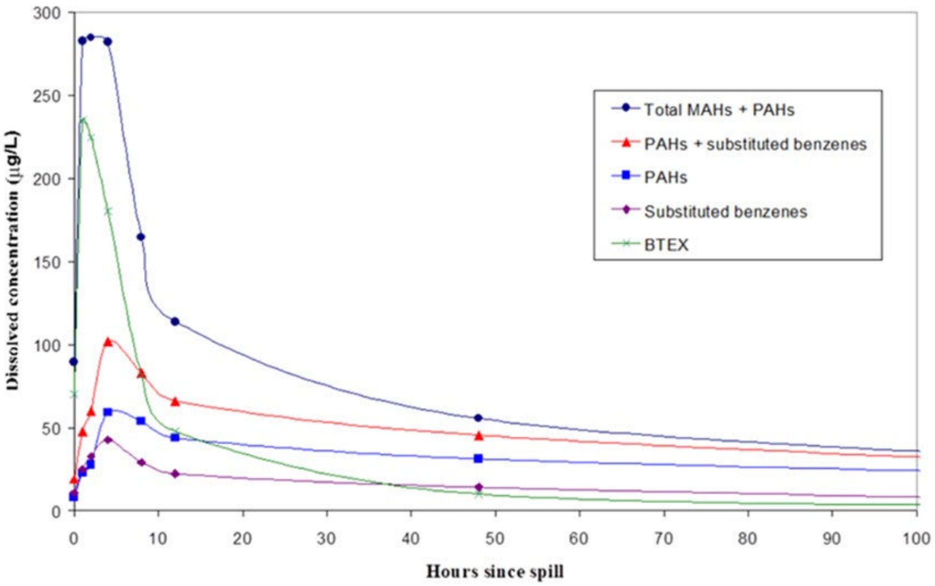

Figure 4. Dissolved concentrations versus time in a tank (data from Payne et al. 1984)

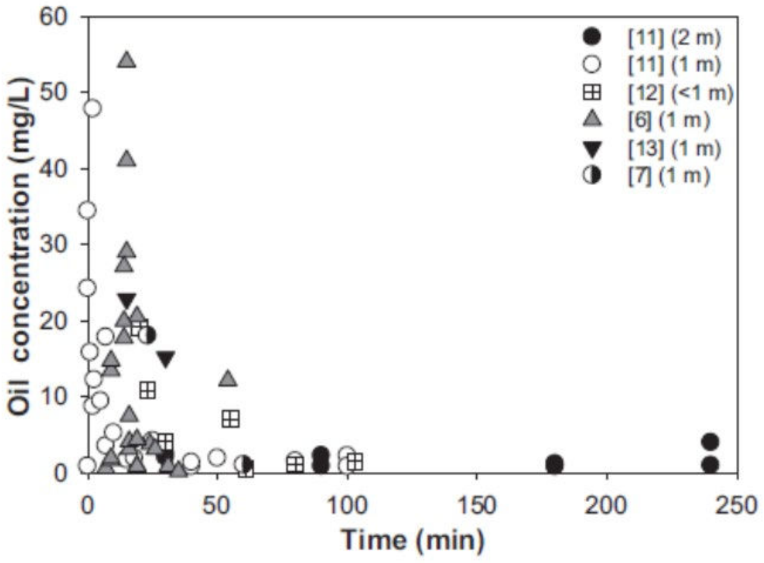

Figure 1. The relationship between measured oil concentrations in the upper water column (≤2 m) versus time from field trials at sea. Data represent oil concentrations from physically ([11], black circles) and chemically enhanced dispersion of oil ([11], white circles; [6,7,12,13]).

Figure 5. Measured oil concentrations versus time after release from field trials (from Bejarano et al. 2014; see source for original citations).

## Organism Behavior

Organisms are not distributed evenly in the environment and are often more concentrated in coastal areas and structural habitats such as coral reefs, rocky reefs, seagrass beds, and seaweeds. Early life history stages of fish and invertebrates are concentrated in these habitats and also in surface waters. In addition, animals move under their own volition and passively (as plankton) in currents, while animals that are attached to structures or otherwise stationary, would be exposed to oil components in plumes that would pass by them. Thus, the duration of exposure to hydrocarbons depends on the distribution of the organisms and their entrainment or movements in relation to the dynamics (changes in) the oil contaminants plume.

A recent modelling study by French-McCay et al. (2024) provides examples of exposure patterns planktonic and stationary organisms might experience for different oil types and spill situations. For example, Figure 6 shows exposures over time for stationary (non-mobile) and planktonic organisms for a modelled 1000 bbl (159 $m^3$) spill of a light crude (Hibernia) in 7 m/s winds.

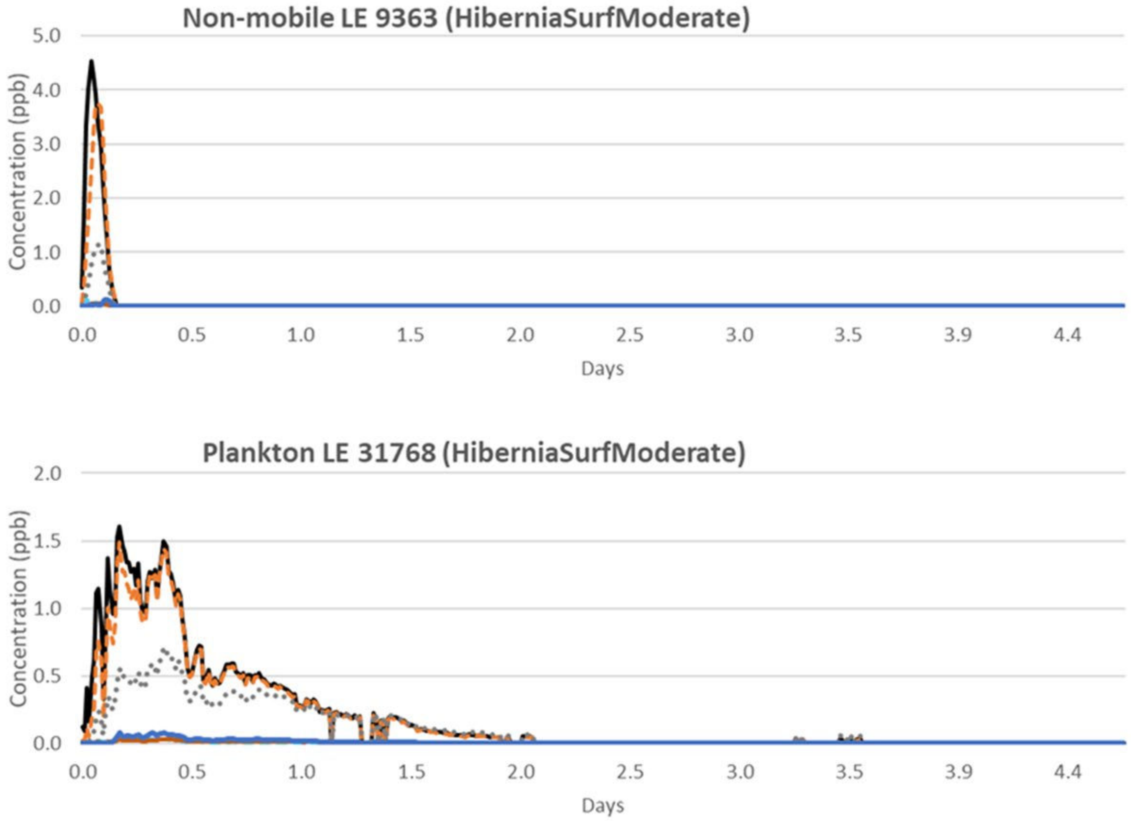

Figure 6. Example exposures to various dissolved oil components over time for stationary (non-mobile) and planktonic organisms after a 1000-bbl spill of light crude.

Note that organisms would need to encounter dissolved concentrations or entrained droplets for any adverse effects to occur, but many are not present in the same depth range as the oil contamination (e.g., surface waters). Other organisms might move through the area of contamination, where their exposure would be for shorter duration than for plankton or stationary organisms. Thus, a 2-dimensional map of maximum concentrations in the vertical dimension, a typical summary presentation of oil modelling results, would not apply to the entire water column. French-McCay et al. (2015a,b) evaluated and modelled vertical and horizontal distributions of fish and invertebrate species and life stages to quantify injuries (French-McCay et al. 2015c) due to the Deepwater Horizon oil spill, taking into account organisms' vertical and other habitat preferences that affected their likelihood and duration of exposure to dissolved oil components.

## Measurements of Oil Concentrations as Total Hydrocarbons

Measurement of the concentration of oil in water, whether in the field or in laboratory studies, is complicated and not all constituents can be identified and measured individually. There are many analytical chemistry techniques used to estimate total hydrocarbons in water, and each of these only measures a portion of the hydrocarbons in oil (Redman et al. 2012; Redman and Parkerton 2015; Yang et al. 2017, Wade et al. 2018; Parkerton et al. 2023a). Recent guidance (Redman and Parkerton 2015; Dettman et al. 2023) recommends measuring BTEX and C5–C9 aliphatics ("volatiles") using purge-trap GC–MS (Gas Chromatograph-Mass Spectrometer), analysis of parent and alkyl PAHs using GC–MS (with target analytes including C0–C4 alkyl decalins and C0–C4 alkyl phenols), and C9–C40 saturate hydrocarbons by GC/FID. Older studies often did not include this detail and range of analyses (see review in Bejarano et al. 2023).

Measurements of C8 to C50 hydrocarbons in oils using these techniques by Yang et al. (2017) identified only 20-30% of the compounds, the remainder of the total petroleum hydrocarbon (TPH) being the unresolved complex mixture (UCM). Mass recoveries (using the presently recommended methods) for gasoline, kerosene, and gas oil are typically >95%, whereas the mass recoveries for heavier substances in crude oils and heavier refined products are often <30%. Mass recoveries of condensates would be somewhat higher than for crude oils but <95%.

The uncharacterized mass (UCM) is due to high-molecular-weight alkanes, asphaltenes, resins, and other large molecules that are either not eluted from chromatographic columns or not well-resolved by gas chromatographic techniques that are available (Redman et al 2012; Yang et al. 2017). In the discussion below THC will refer to the true concentration of total hydrocarbons (and related compounds), whereas TRH will be used to refer to the measured (i.e., recoverable) portion of the THC.

As only some of the compounds in oil are measurable individually, and because of the desire to be inclusive of effects from hydrocarbons in oil droplets (e.g., the entrained droplets in SIMAP model outputs) as well as the dissolved phase, aquatic toxicologists have attempted to use TRH as a metric for evaluating toxicity of oil exposures. However, given that TRH does not actually measure THC and the complexities described above, among others related to experimental design and conditions, development of lethal toxicity values (e.g., LC50s) or sublethal thresholds based on total hydrocarbons (THC) is problematic. Furthermore, the hydrocarbon compositions in exposure media are highly variable from experiment to experiment, leading to estimated toxicity thresholds that range over many orders of magnitude, as is evident from reviews of these data (e.g., see NRC 2003; NASEM 2020).

## Oil Bioassays

Toxicity studies often have used experimental systems containing media preparations made by mixing oil in water long enough to bring the composition of oil components into equilibrium, i.e., Water Accommodated Fractions (WAFs) of oil. WAFs are purposely static, and thus, do not represent variable field compositions and changing concentrations. WAFs can be made with fresh or weathered oil, but bioassays using a particular WAF do not account for weathering changes in the field. Further, as the concentrations of individual compounds in WAFs change from test to test and the individual compounds vary in toxicity, effects levels based on total PAH or TRH are highly variable. LC50s or other toxicity metrics based on nominal additions of oil to media (in an attempt to measure a THC effects level) are unreliable because some compounds evaporate, adsorb to test containers, bioaccumulate, and degrade in test chambers, and the oil component mixture composition is unknown (Hodson et al. 2019).

The 2023 Special Issue of Aquatic Toxicology (Loughery et al. 2023; Stubblefield et al. 2023; Alloy et al. 2023; Parkerton et al. 2023a,b; French-McCay et al. 2023; Dettman et al. 2023; Bejarano et al. 2023; Lee et al. 2023) contains recommendations for oil toxicity test methods, including reviews of the issues summarized here. Loughery et al. (2023), Stubblefield et al. (2023), Alloy et al. (2023), Parkerton et al. (2023a,b), Dettman et al. (2023), Bejarano et al. (2023), and Lee et al. (2023) provide guidance for aquatic toxicity studies, building on and updating the standardized toxicity test protocols developed by the Chemical Response to Oil Spills: Ecological Effects Research Forum (CROSERF; Aurand and Coelho 2005, Singer et al. 2000).

Thus, toxicity data developed using WAFs should not be applied directly to field conditions to evaluate effects. TU modelling should be used to evaluate the toxicity in the bioassay mixture (Parkerton et al. 2023a), and the toxicity of the mixture in the field, for which the composition and toxicity will differ. Whole oil bioassays using WAFs or other preparations exposing organisms to dissolved components and entrained oil are useful and important for determining species and life stage sensitivities and for validating the predictions of toxicity models based on additive effects of oil components (French-McCay et al. 2023).

WAF-based effects levels have been used to develop concentration thresholds above which effects might be expected. Note that even if conditions in the field could be considered quasi-steady state (i.e., in an enclosed quiescent water body where oil dilution is prevented by geography and where weathering is slowed by environmental conditions and isolation from the atmosphere) and the experimental oil composition of a test were representative of the field under the specific conditions tested, the experimental effects level would over-estimate toxicity to the extent it is applied to modelled oil, as the model accounts for the entire mass of oil, as opposed to what was measurable in the test.

## Experiments Exposing Organisms to Oil Droplet Dispersions

### Exposure Methods Produce Long Steady State Exposures, Negating Weathering and Dilution

The Norwegian studies (Nordtug et al., 2011b; Olsvik et al., 2011, Hansen et al. 2012, 2015, 2016, 2018, 2019a; Sørhus et al. 2015, 2016, 2017, 2021a,b, 2023a,b; Sørensen et al. 2017, 2019) exposed early life stages of fish (eggs, embryos and larvae of cod and haddock) to dispersions of oil micro-droplets (<60 microns in diameter; e.g., mean volume based diameter in the range of 10–16 um; Nordtug et al., 2011b; Hansen et al. 2017, 2019a,b) supplied via a continuous dosing system described in detail in Nordtug et al. (2011b). They used artificially weathered oil where the oil was distilled to about 150-200°C (Nordtug et al., 2011b; Sorensen et al. 2017; Hansen et al. 2019a,b, 2025), enough to remove substantial amounts of the BTEX and other volatile organic carbon (VOC) compounds but not the naphthalenes (2-ring PACs) and 3+ ring PACs (as evidenced by chemical analyses, e.g., Hansen et al. 2019a,b). The oil dispersions used in the exposures were prepared continuously; thus, the exposures were to relatively fresh oil, still containing naphthalenes and 3+ ring PACs. Typically, exposure times to these dispersions were 4 or more days (e.g., review table in Hansen et al. 2025). For example, Sorensen et al. (2017) exposed embryos to essentially constant concentrations of freshly prepared oil dispersions starting at 1 day post fertilization (dpf) lasting until hatching of the larvae at 11 (haddock) or 12 (cod) dpf (i.e., for 10-11 days).

This approach using the oil droplet generator (ODG) to continuously dose the experimental media of a toxicity test provides relatively constant exposures to oil dispersions. Hansen et al. (2025) describes it as such and with a goal of reaching equilibrium between the oil dispersion in the media and the test organisms. While this approach provides a well-controlled system to evaluate toxic effects and is valuable for the purpose of studying mechanisms and validating toxicity models, it does not represent conditions in the field during an actual oil spill. Concentrations of oil droplets and dissolved constituents after oil spills into water bodies are not ever steady-state or in equilibrium between the dissolved and particulate phases or with aquatic organisms. Thus, effects levels derived in such an ODG system, expressed as total PAH or total PAC concentrations, should not be used as thresholds in risk assessments, particularly not for oil droplets. Data from these tests can and should be used to identify mechanisms, evaluate bioavailability, and test and verify toxicity models, such as performed by Hansen et al. (2019b).

In an open water system, most of the naphthalenes and a substantial fraction of the 3-ring PACs typically dissolve in a few hours (surface releases) to a few days (deepwater blowout) from oil micro-droplets ~100 μm in diameter (French-McCay et al. 2021b, 2024) and faster from smaller droplets such as those used in the continuous dosing system experiments. Figure 7 shows the loss over time of soluble and semi-soluble pseudo-components (as defined in French-McCay et al. 2021a,b) by dissolution from 50-micron droplets, a size at the high end of the range of droplet sizes used in the experimental studies. The AR5 to AR8 pseudo-components comprise the PACs, with log(Kow)s of AR5, AR6, AR7 and AR8 being 4.1, 5.1, 5.1 and 5.6, respectively (which are indicative of their dissolution rates). Chemical measurements of samples from subsurface waters taken during the Deepwater Horizon oil spill showed that the droplets were highly weathered and contained much lower concentrations of PACs than fresh or artificially weathered MC252 oil (Payne and Driskell 2017, 2018; Driskell and Payne 2018).

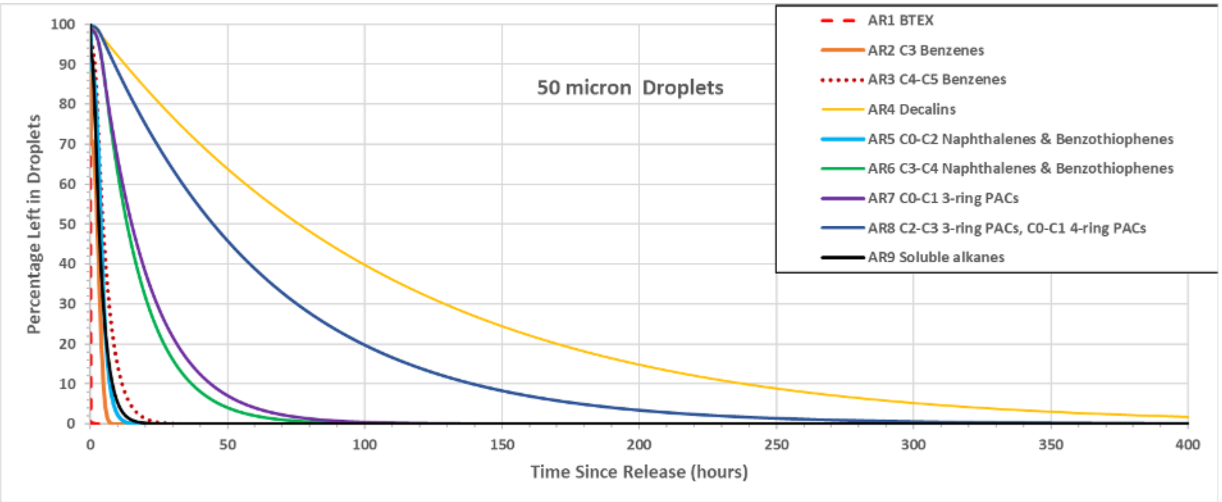

Figure 7. Weathering of 50-micron oil micro-droplets of MC252 light crude oil showing loss of soluble and semi-soluble pseudo-components (as defined in French-McCay et al. 2021a,b) by dissolution over time.

In experimental studies using continuously supplied dispersions of freshly prepared oil micro-droplets mostly <20 um in diameter, dissolution of 2-3 ring PACs would occur throughout the entire exposure period. The dissolved phase would contain relatively high concentrations of PACs throughout the tests. Thus, the exposures tested were not representative of a real-world spill condition (except potentially at the source location of a continuous release). Furthermore, the exposure periods were 4-10 days in confined containers. In addition to negating the effect of weathering of the micro-droplets, dilution was also prevented.  Thus, the experimental exposure times to dissolved components were much longer than would occur in waters where dilution would rapidly reduce concentrations. In areas away from the source, not only would droplets and initially dissolved components be diluted, but the droplets would also be already depleted of the soluble and semi-soluble components.

## Effects Thresholds for Oil Dispersions Include Dissolved PACs

Carroll et al. (2022) in their review of oil dispersion effects levels stated that the lowest reported effect concentration was 10 µg/L of oil dispersion (containing 0.1 µg/L PAH; Cresci et al., 2020). Cresci et al. (2020) had found that Atlantic haddock larval swimming speeds were less variable and 31-40% slower than controls after 8 days of continuous exposure to fresh oil dispersions prepared as described in Nordtug et al. (2011b). By using their in-situ chamber and this approach, Cresci et al. (2020) forced continuous exposure to fresh oil for 8 days, which did not lead to environmentally relevant concentrations as claimed. The system restricted both weathering (dissolution from) and dilution of the oil droplets. The chemistry measured whole water samples from the chambers, and so included PACs both dissolved and in the droplets. Thus, this sublethal effect concentration of 10 µg/L should not be applied to concentrations of entrained oil droplets of any size, whether considered inclusive of associated dissolved components or as distinct from dissolved concentrations.

Hansen et al. (2012) reported LC50 values of 16.06 (13.53-19.07) µg/L total PAH for Calanus finmarchicus exposed to mechanically dispersed oil. Carroll et al. (2022) stated: "The lowest PAH effect concentration is 1.34 µg/L ∑PAH (Hansen et al., 2016)." Hansen et al. (2025) does not report lower effects levels but does report PAH body burdens at somewhat lower concentrations. In all these studies, the exposures were for 4 days to continuously supplied oil dispersions, which include both dissolved and droplet-bound PAHs. Sørhus et al. (2023a) recommended a 0.1 total PAH threshold for risk assessments based on observed acute and delayed mortality at embryonic, larval, and juvenile stages after 8-day exposures to 2.2, 0.39, and 0.27 µg total PAH/L, respectively, using the Nordtug et al. (2011b) ODG method to create oil dispersions. In addition to the exposures being unrealistically long to relatively un-weathered oil, in all these studies, the measurements of PAHs were of whole water samples, i.e., including the dissolved compounds. In open waters droplets and dissolved compounds would separate due to the buoyant rise of droplets. Thus, these should not be translated to equivalent entrained oil droplet concentration thresholds.

Results of the modeling analysis by Hansen et al. (2019b) indicated that dissolved phase oil primarily dictated observed toxicity for survival and growth of cod larvae, not dispersant nor droplet oil, consistent with earlier studies (e.g., Carls et al., 2008; Nordtug et al., 2011b; Olsvik et al., 2011). Effects levels from 4-day exposures expressed as total PAHs were 6-36 µg total PAH/L. Observed effects were found to be successfully described using dissolved phase Toxic Units (TUs) for experiments with and without droplet oil. Total PAHs were found to account for 10% or less of the predicted TUs for the artificially weathered naphthenic oil investigated. Unresolved (i.e., not measured by gas chromatography–mass spectrometry, GC/MS) aromatic and naphthenic aromatic components contributed the majority of TUs (Hansen et al. 2019b). Thus, expressing the toxicity values as 6-36 µg/L of total PAH underestimates the actual concentrations of toxins causing the observed effects, biasing low a total-PAC threshold based on this or similar experimental data.

Nordtug et al. (2022) conclude, with respect to these experimental studies with the continuous oil dispersion dosing system: "From a methodological point of view, the observed droplet associated toxicity could be explained by retainment of water-soluble components during the filtering procedure used to generate the WSF, i.e., the WSF composition before and after filtration are not identical. Alternatively, the water phase of the dispersions may not have been in equilibrium before the separation of the WSF through filtration (Hansen et al., 2019a). … The third possibility is that even in a dispersion at equilibrium, the oil droplet will act as a reservoir or buffer constantly replacing components that are bioaccumulated to maintain a slightly higher WSF over time. Finally, the droplets attached to the eggs may cause uptake directly from the oil into the tissue, as seen in the study by Sørensen et al. (2019). All these factors will contribute to an overestimation of the toxic effect of oil droplets. As we are not able to quantify the potential impacts of these confounding factors, we conclude that it is plausible that the oil droplets contribute to the oil toxicity observed in the current experiment, but the mass-related contribution is much lower than for the water-soluble fraction."

## Modelling Study by Frøysa et al. (2025)

The objectives of Frøysa et al.'s (2025) modelling analysis were to answer these questions for their example scenario intended to simulate potential exposures to Atlantic haddock eggs off the coast of Norway:

1. What is the relative contribution to oil spill exposure of fish eggs from oil droplets and dissolved oil?
2. Is the impact region of oil droplets larger than that of dissolved oil?
3. Do oil droplets adhering to eggs lead to prolonged exposure durations as compared to dissolved oil?

Frøysa et al.'s (2025) model assumed that oil micro-droplets were released at the surface continuously at a rate of 4,500 $m^3$/day (1,188,774 gal/day = 28,304 bbl/day) for 90 days. This release rate is of the same order of magnitude as the Deepwater Horizon oil spill (Lehr et al. 2010). Droplets were assumed 2-59 μm in diameter, accounting for 30% of the spilled mass, which is not realistic for a surface release (which could not occur at this rate for 90 days) or a blowout from the sediments at the modelled location (<100 m deep). The model scenario did not account for the energy and nearfield dynamics of the release. The nearfield model (Johansen 2003) built into the OSCAR model (Johansen et al. 2013) utilized in the SYMBIOSES system (Carroll et al., 2018, 2022, 2023) would show that most of the oil would be released in large droplets millimeters in diameter, as would the Li et al. (2017) model developed based on essentially the same experimental dataset as used by Johansen et al. (2013). The assumption that all the droplets modelled were <60 μm in diameter caused them to remain suspended in the surface waters indefinitely, whereas droplets >100 μm would surface (and those >500 μm, as would be typical for a surface release, would surface within minutes from surface water). Indeed, SINTEF's DeepSpill experimental oil releases from 844 m (Johansen, 1984, 2003; Johansen et al. 2003) resulted in millimeter diameter oil droplets surfacing within a few hours.

Frøysa et al.'s (2025) model did not consider oil composition as described by the OSCAR model pseudo-components, although they do admit this limitation by noting a "distinct difference between dissolved oil and oil droplets is the oil composition". Exposure was recorded as to total dissolved and total oil. Frøysa et al. (2025) stated: "Our modeling results indicate that oil droplets play a larger role in exposure than dissolved oil, in having both greater mass concentrations and covering larger areas." However, oil concentration of 1 µg /L as a droplet would not be equivalent in toxicity to 1 µg /L of (typically measured) dissolved aromatics, which would be primarily PACs in bioavailable form. The oil droplets were <60 um in diameter, so the dissolution would occur very quickly (Figure 7), depleting the soluble and semi-soluble compounds (including the PACs) from the micro-droplets, leaving residual compounds that would not be bioavailable via dissolution from the droplet stuck to an egg and uptake into an egg across its membrane(s). One needs to consider weathering state (i.e., composition) of the oil droplets to evaluate potential contribution to toxicity. A Toxic Unit type approach should have been applied to compare the toxicity of the droplets versus the dissolved. Also, uptake into the eggs and total mass balance should be part of the accounting.

Frøysa et al.'s (2025) findings included that the mean exposure duration for suspended droplet oil was shorter than for dissolved oil, in agreement with findings by French-McCay et al. (2024). By assuming 100% adherence of micro-droplets with eggs, Frøysa et al. calculated long exposure durations to droplets. However, Frøysa et al.'s (2025) conclusion that "impact assessments not taking oil droplets into account may drastically underestimate the impact of oil spills on the marine environment" is hypothesized and is not supported by published experimental work, their modelling study, or other modelling analyses. Furthermore, the droplet adherence rate in Frøysa et al.'s (2025) model was about 40 times as high as observed in experiments by Hansen et al. (2018) with similar (high) exposure concentrations to the model's assumed exposures. The compositional changes and decrease in bioavailable toxins that would occur in micro-droplets over time was not considered. As noted, the simulated oil spill was not realistic, with far more mass released as micro-droplets than possible in the Norwegian Sea or any other location in <100 m depth.

Thus, with respect to the questions posed by Frøysa et al. (2025):

- What is the relative contribution to oil spill exposure of fish eggs from oil droplets and dissolved oil? This cannot be answered using the results of their modelling study.
- Is the impact region of oil droplets larger than that of dissolved oil? This also cannot be answered using the results of their modelling study. Evidence from experimental studies and other modelling work indicates that this is not the case.
- Do oil droplets adhering to eggs lead to prolonged exposure durations as compared to dissolved oil? The answer depends on what is considered. Exposure of sticky eggs to residual non-bioavailable portions of the oil could be prolonged by adherance of droplets to egg surfaces. However, this modelling study and experimental data do not support the assertion that exposures to oil droplets could be more impactful than via the dissolved phase.

Thus, the effects of micro-droplet oil on aquatic biota are mainly due to the increased surface area of the oil droplets (as compared to floating oil or large droplets) facilitating dissolution of soluble and semi-soluble components and bioavailability to aquatic biota.

## Recommendations for Thresholds

### PAH-Based Threshold for Dissolved Hydrocarbons

Because most of the toxicity from dissolved hydrocarbons and related compounds is due to PAHs and related compounds, particularly in surface waters, PAH-based lethal and sublethal thresholds are typically used to evaluate the risks of oil contamination. In many bioassays, measured total PAH concentrations have been used to quantify toxicity endpoints, with the understanding that the measured PAH concentrations reflect the effects of any dispersants applied (which increase concentrations in the water, Bejarano et al. 2014) and other unmeasured constituents associated with the PAHs are contributing to the observed toxicity (Bejarano et al. 2017; Forth et al. 2017; Parkerton et al. 2023a), although with less effect (French-McCay 2002).

PAH concentrations on the order of tens to hundreds of micrograms per liter (µg/L, i.e., ppb) have been shown to be acutely toxic to aquatic biota (French-McCay 2002, 2016). French-McCay (2002) reviewed available laboratory oil and PAH acute bioassay data, finding that 95% of species and life stages exhibited 50% mortality between 6 and 400 ppb (µg/L) total PAH concentration if continuously exposed for >96 hours. Based on this result, the 6-ppb total PAH value should be protective of 97.5% of species and life stages exposed for >96 hours. Early life history stages of fish appear to be more sensitive than older fish stages and invertebrates. Bioassay data compiled by French-McCay (2002) indicate that 96+ hour LC50s of juvenile and adult fish are >100 ppb. Bejarano et al. (2017) developed species sensitivity distributions (SSDs) for acute bioassay tests using WAFs involving typically 4-day exposures. Bejarano et al (2017) found that the 5th percentile sensitivity (Hazardous Concentration for 5%, HC5, Posthuma et al. 2002, which is protective of 95% of species and life stages) for lethality ranged from 28 to 282 ppb total PAH concentration for fish and invertebrates from a wide variety of latitudes and habitats, a range similar to that predicted by French-McCay (2002). Note that in recent years the 5th percentile for a range of species sensitivities in toxicity tests has come to be used in practice for developing thresholds, and this is termed the HC5 (Barron et al. 2013; Fox et al. 2021; Stubblefield et al. 2023). Thus, for acute lethality and to the nearest order of magnitude (given uncertainties), total PAH LC50s (for >96 hours of exposure) generally range from about 10 ppb for sensitive early life history stages to 300 ppb or more for less sensitive species and older life stages.

Sublethal effects may occur at lower concentrations. For PAHs, USEPA (2003, 2008) has derived and used a factor 10 to estimate a sublethal effects threshold (Final Chronic Value, FCV, or Predicted No Effect Concentration, PNEC) based on a lowest acute toxicity-based endpoint such as the HC5 based on 96-hour LC50 tests. In a recent study by McGrath et al. (2018), the acute-to-chronic ratio (ACR) for hydrocarbons was found to average 5.22 (range 1.7-12 for the 10th and 90th percentiles of the observations, including consideration of one high outlier), therefore, the factor 10 is conservatively protective. Thus, the sublethal effects threshold (PNEC) for PAHs would be 1 µg/L (ppb) where exposure durations are sufficiently long (i.e., days to weeks) for such effects to occur. For oil spills in open water, the acute threshold of 10 ppb would be appropriate, given short-term exposures that occur in marine and coastal waters. Further details are available in French-McCay (2009) and French-McCay et al. (2018). These total PAH thresholds have been used in many environmental risk assessments (e.g., French McCay et al., 2005, 2012, 2018; NOAA, 2013).

In addition, photo-chemical reactions upon exposure of oil or contaminated organisms to sunlight produces photoproducts (new compound structures, by photo-modification) or free radicals in organisms (photosensitization to activated PAHs and related compounds) that could adversely affect exposed aquatic organisms. Photosensitization can occur in early life stages of fish and invertebrates that are translucent or transparent and vulnerable since they are undergoing development (Alloy et al. 2023). Specific thresholds have not been developed that account for photo-toxicity. However, studies reviewed by Alloy et al. (2023) indicate an effects threshold accounting for phototoxicity would be approximately 1 µg/L (ppb) . Exposure durations to both PAHs and light would need to be sufficiently long (i.e., days or weeks) for such effects to occur. Further details are available in French-McCay et al. (2018) and Alloy et al. (2023). The adoption of a threshold of 10 ug/l (ppb) for an instantaneous exposure is suitably conservative given that orders of magnitude higher concentrations would be required to produce the same effects over an instant.

### THC-Based Threshold for Entrained Droplets

The concentrations of entrained droplets output by the SIMAP model represent hydrocarbons and other compounds that are mostly not bioavailable due to the weathering (volatilization, dissolution, and degradation) accounted for in the model. The soluble and semi-soluble fractions dissolve from the droplets over time, and a potential effects analysis based on the dissolved hydrocarbons characterizes their risk. However, to satisfy concerns that entrained droplet hydrocarbons might infer some risk, THC-based thresholds have been used.

In addition to considering total PAHs, Bejarano et al. (2017) developed SSDs using *measured* TPH (i.e., TRH; aromatic and aliphatic hydrocarbons [C9-C44], which includes parent and alkylated homologue PAHs and/or parent naphthalene) as a metric. The range of HC5s was found to be 1 to 5.6 ppm (mg/L) as TRH for WAF preparations from crude oils with API densities ranging from 25 to 44. The HC5 for light refined Group 2 fuel oil (API 35 – 45) was 0.5 mg/L TRH. The TRH HC5 decreased with higher API (lower oil density), i.e., with oils that have a higher percentage of light hydrocarbons (C9-C44) including PAHs. WAFs included aqueous exposure media prepared by physical (low energy water accommodated fraction, LEWAF; and moderate energy WAF or MEWAF) or chemically enhanced oil dispersion (chemically enhanced water accommodated fraction, CEWAF). Note that the TPH measurements were on WAF media, which are preparations that purposely select for the soluble hydrocarbons and remove the insoluble larger hydrocarbons in the oil (Redman and Parkerton 2015). Thus, the TPH measurements of the WAF media are on a portion of the oil, not the full oil (see also Parkerton et al. 2023a for further detail).

This limitation also exists for measurable TPH (i.e., TRH) thresholds presented in Smit et al. (2009), which has been used as a proxy THC threshold for oil spill modelling in the past. Furthermore, the thresholds developed in Smit et al. (2009) are for no-observed-effect concentrations (NOECs) based on sublethal effects of chronic exposures of 7 to 183 days (e.g., the lowest test NOEC was for a 33-day exposure). As discussed above (and in French-McCay et al. 2023), such long exposure times do not occur in open (marine and coastal) waters after oil spills and effects concentrations are much higher for short-term exposures.

The ANZECC 2000 guidelines and derivation of a TPH-based sublethal threshold include a review of a few studies available at the time but depend on the analysis by Tsvetnenko (1998) for a threshold. Tsvetnenko (1998) followed US EPA methods (Stephan et al. 1985; US EPA 1994) to develop SSDs and HC5s but did not recognize that TPH measurements of Water-Soluble Fractions (WSF) and WAFs do not represent the same composition as the THC in the field. They are biased towards a mix of soluble and semi-soluble components that are the toxic components of the oil (as explained above). Tsvetnenko (1998) developed a lethal HC5 for TPH of 168 µg/L (ppb) and then applied an ACR of 25 to obtain a sub lethal threshold of 7 µg/L (ppb) as TPH. Note that recent reviews do not support this ACR (McGrath et al. 2018). An ACR=10 would be sufficiently protective for chronic exposures. Even if it were assumed that TPH measurements on WSF/WAF media were representative of THC (which they are not), the HC5 of 168 µg/L should be corrected (upward) for the fraction of the THC of the source oil actually measured by the TPH analyses used and should not be corrected (downward) for the volatilization loss (in table 8.3.24, ANZECC 2000 appears to have lowered the measured TPH toxicity values by a factor of 10, based on Hamoda et al. 1989). The needed data are not available, but likely the correction would be an order of magnitude, given the time and nature of those TPH measurements and apparent volatilization in the experiments. However, it is more appropriate to use recent data where better analytical methods were used, if THC is to be the basis of a threshold.

Because PACs (typically referred to as PAHs) are the most toxic components of oil and crude oils typically contain about 1-3% PAHs by mass (French-McCay 2002; Forth et al. 2017), the entrained oil threshold expressed as total hydrocarbon concentration (THC, not TPH) should be at least 330-1000 µg/L (330-1000 ppb) for *fresh* crude oil, corresponding to the dissolved threshold of 10 ppb (which is based on total PAHs). However, as oil weathers, PAHs are lost to volatilization, dissolution, and degradation and the ratio of the entrained to dissolved threshold should be higher than this range. Thus, the whole-oil threshold of 1,000 ppb (1 ppm) is conservative (low, and highly protective of aquatic resources).

Based on the review of toxicity studies by Bejarano et al. (2017), THC lethal effects levels of 28-300 mg/L (28-300 ppm) were found for a range of crude oils for species from all geographical areas globally. An exposure concentration of 1,000 ppb (1 ppm or 1 mg/L) of measurable TPH was deemed a low level of concern for sensitive life stages in marine organisms by Kraly et al. (2001). In reviews by NRC (2005) and NASEM (2020), 1,000 ppb (1 mg/L) was found to be at the low end of the range where sub-lethal impacts from acute exposure have been observed.

Correcting for fraction measurable, this indicates a THC-based threshold of 3,000 ppb would be appropriate for modelled entrained crude oil. Thus, a THC threshold of 3 mg/L (3 ppm; 3,000 ppb) is an appropriate threshold for crude oils for use in risk assessments and for defining an EMBA. For light distillates and condensates, 1 mg/L (1 ppm; 1,000 ppb) is an appropriate threshold.

Notably, NOPSEMA (2025) has recommended use of a 1 mg/L (1,000 ppb) threshold as their exposure value high. NOPSEMA (2025) states (in their Table 1): “It represents a concentration of entrained oil at or above which there is an increased likelihood of consequences on ecological, social, economic and cultural features of the environment. For example, sublethal effects to sensitive species or life stages may occur at or above this concentration (e.g. fish and invertebrates) (French-McCay, 2025; Negri et al, 2025).” NOPSEMA (2025) also recommends an order of magnitude lower value (exposure value moderate) of 100 ppb as “a conservative exposure value to account for the uncertainty in consequences on ecological, social, economic and cultural features of the environment.” Given that a 1 mg/L (1,000 ppb) threshold is already conservatively low, protective of exposures to un-weathered fresh oil for weeks or longer, and the use of stochastic modeling to characterize uncertainties in where oil might be transported, inclusion of this extra uncertainty factor is overly conservative.

### Consideration of Duration of Exposure for Developing Thresholds

The use of LC50s for >48 hours of exposure, or chronic endpoints for longer exposure times, as instantaneous thresholds for oil spills is highly conservative. Acute aquatic toxicity thresholds would be sufficiently conservative for oil spills in marine and other open waters (as opposed to ponds or other contained systems). There is no need for an ACR correction for evaluating acute toxicity to aquatic biota from oil spills in open waters.

In the ANZECC 2000 guidelines, no observable effects concentrations (NOECs) after long exposure durations are recommended. This is highly protective for chronic steady state concentrations of hydrocarbons. Again, since oil spill water column exposures are acute, and durations of exposure are for hours and not days, acute toxicity data (e.g., Final Acute Value, FAV) should be used and there should be no ACR conversion.

To in some part compensate for the effect of exposure duration on effects levels, a higher threshold appropriate for short duration exposures could be used. Alternatively, the modelled exposure concentrations could be time averaged over 1 to several hours.

Another approach would be to use a dose metric as a threshold. The LC50s quoted above are typically for 96 hours of exposure. Thus, the 10-ppb dissolved PAH threshold would be equivalent to 960 ppb-hours of exposure (i.e. 10 ppb average concentration persisting for 96 hours). The 1,000 ppb THC threshold would be equivalent to 96 ppm-hours. Model results could be integrated to calculate the ppb-hours of exposure, therefore addressing the duration of exposure issue.

## Conclusion

### Recommended Entrained Oil Thresholds

For the dissolved hydrocarbons, the 10 ppb PAH threshold is conservatively protective of aquatic biota considering acute effects from short-term exposures that could occur following oil spills in ocean waters. If THC in oil droplets is to be evaluated as an environmental risk, 1 mg/L (1,000 pp; 1 ppm) would be sufficiently conservative for oil droplets of all oil types and all weathering states, with 3 mg/L (3,000 ppb; 3 ppm) an appropriate threshold for crude oils. For light distillates and condensates, 1 mg/L (1,000 ppb; 1 ppm) is an appropriate threshold.

In confined water bodies, such as rivers or estuaries, where dilution is restricted, exposure durations to droplets could last longer than in open water (i.e., open coastlines and seas). However, oil droplets would weather over time such that the soluble and semi-soluble components would dissolve out of the droplets leaving oil residuals with low bioavailability. This weathering would occur much more quickly for small droplets produced in turbulent conditions (such as high waves). If fresh oil enters confined waters, they could produce dissolved concentrations that potentially could adversely affect aquatic biota if in sufficient concentrations, given the longer exposure times resulting from slower dilution.  Such a situation did occur during the *North Cape* oil spill (French-McCay 2003). As demonstrated by that model validation analysis of the *North Cape* oil spill, it was the dissolved concentrations that adversely affected aquatic biota, not entrained droplets, as they were rapidly spent of their PAC content . The risk of such outcomes would be addressed directly by assessment of the dissolved concentrations during risk assessment modelling. It cannot be correctly inferred from calculations for entrained oil.

### Limitations of Using Screening Thresholds

When using thresholds, it should be noted that toxicity is determined by the specific mix of hydrocarbon concentrations in the exposure medium and the toxicity of each component. Use of a THC threshold does not address the complexities of the mixture of hydrocarbons of various toxicities. For these reasons, recent evaluations of the aquatic effects of oil have focused on the most toxic and bioavailable components (i.e., dissolved hydrocarbons) and have not used measured TPH or THC as a metric. The focus of a risk assessment should be on the dissolved hydrocarbon exposure, along with floating oil and shoreline oil, as opposed to entrained oil droplets. The review series (Loughery et al. 2023; Stubblefield et al. 2023; Alloy et al. 2023; Parkerton et al. 2023a,b; Dettman et al. 2023; Bejarano et al. 2023; Lee et al. 2023; French-McCay et al. 2023) provides guidance for aquatic toxicity studies and their application to risk assessments and modelling-based analyses.

It should be recognized that the use of potential effects thresholds to summarize oil spill model results from a dynamic model such as SIMAP is a simplified approach that cannot address all the complexities of oil exposures and effects occurring in the field (Hook 2020; French-McCay et al. 2023). French-McCay et al. (2023) review a range of approaches (i.e., aquatic toxicity modelling tiers), with increasing complexity and data requirements, along with accompanying uncertainties, to address the varying needs of spill responders and risk/impact assessors. At the simplest level (Tier 1), modelling is used to estimate maximum exposure concentrations of oil or some fraction of oil at any time after a spill. These spatially defined concentrations are compared to screening thresholds to evaluate areas or volumes at risk for potential adverse effects. However, threshold approaches cannot readily address the varying composition of oil as it weathers, differing bioavailability via the dissolved phase, or changing effects levels with duration of exposure. Tier 2 and Tier 3 modelling approaches consider the dynamics of oil movements and duration of exposure that would be experienced by aquatic biota, while still being based on an assumed representative mixture of components causing effects. A Tier 4 modelling approach considers the changing composition of the oil components using a TU approach, which could be focused toward effects on sensitive species and life stages. Population level effects modelling called for by Hook (2020) among others would require detailed modelling of species and life-stage specific uptake, metabolic, biotransformation, and elimination processes, along with effects on survival, growth and reproduction. Considering ecosystem level effects would require considering multiple species and interrelationships between species and the habitats they occupy in a mechanistic manner, as not all situations could be experimentally evaluated or monitored in the field. The feasibility and uncertainties of the more detailed higher tier approaches need to be considered when planning any assessment.

### Considerations Regarding Defining EMBAs and Operational and Scientific Monitoring Planning Areas

It should be noted that exceedance of threshold does not infer adverse effects would occur. While this has been understood by NOPSEMA (2025) and others, if a threshold is overly conservative, use of the EMBA or Operational and Scientific Monitoring Planning Area delineation would not focus monitoring on locations where there is a potentially high risk of consequences. Near the outer bounds of an area defined by an entrained oil threshold of 10 µg/L (~10 ppb, as recommended by NOPSEMA 2025), there is a very low probability of concentrations exceeding 10 ppb, the exposure durations are typically minutes to a few hours, and the oil is highly weathered (and so less or not toxic). Thus, a more realistic threshold would be appropriate, and it need not be protective of every single individual organism that might possibly encounter one droplet of entrained oil, regardless of likelihood or its ability to cause any adverse effect.

Monitoring should focus on locations most at risk of consequences, as opposed to those with marginal, if any, risk of consequences. Such locations include near the release site, in shallow waters where oil could accumulate, in sensitive habitats, and in areas where there are species with protection status. Monitoring to measure any change is challenging even for very large spills due to natural variability of marine habitats and populations, and of water quality (including the influence of natural oil seeps) and the large volumes of oceanic waters and habitat areas that would need to be sampled (as noted by Fodrie et al. 2014, Hook 2020, and other authors). Sampling effort, experimental design and detection limits of monitoring tools, including analytical chemistry capabilities, need to be carefully considered, otherwise resources will be wasted. The application of potential effect areas, defined by the NOPSEMA (2025) recommended low thresholds as instantaneous occurrences, to plan for scientific monitoring is problematic because those thresholds will be below detection, are recognized as being lower than are expected to cause any measurable ecological effects, and, being instantaneous, will be highly unlikely to coincide with the timing of sampling via monitoring in the field. Directing monitoring to areas where exposures are low and ephemeral, hence where detection would be difficult if not infeasible, would be highly likely to waste resources (and available time before effects have passed) and to report no effects, compared to directing resources to other areas subject to higher concentrations for longer.

Thus, monitoring should be planned for where higher thresholds than those recommended are expected to increase the potential to successfully detect change or effects. The exposure value high of 1,000 µg/L (~1,000 ppb, recommended by NOPSEMA [2025], which is based on potential for sublethal effects to sensitive species or life stages that may occur at or above this concentration, e.g. fish and invertebrates [French-McCay, 2025; Negri et al, 2025]) would be an appropriate delineator for monitoring for adverse effects. The exposure value moderate of 100 µg/L (~100 ppb, recommended by NOPSEMA [2025], which is based on a "conservative exposure value to account for the uncertainty in consequences on ecological, social, economic and cultural features of the environment", is very conservative and in practice such low concentrations and any resulting effects would not be feasible to measure. Experience during the response to the Deepwater Horizon oil spill (Boehm et al. 2016; Wade et al. 2016; Driskell et al. 2018; Payne and Driskell, 2017, 2018; French-McCay et al., 2021b), the most sampled of any oil spill in history, demonstrated how intractable it would be to undertake such a monitoring program as is recommended by NOPSEMA (2025).

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